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EUROPEAN PATENT APPLICATION

② Application number: 89830122.1

⑥ Int. Cl. 4: **C 07 D 235/28**
A 61 K 31/415, C 07 D 235/02

③ Date of filing: 17.03.89

④ Priority: 25.03.88 IT 1985888

④ Date of publication of application:
27.09.89 Bulletin 89/39

④ Designated Contracting States:
AT BE CH DE ES FR GB GR IT LI LU NL SE

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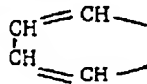
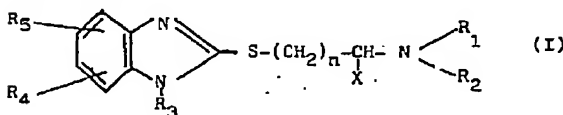
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④ Pharmacologically active alkylthiobenzimidazole derivatives and process for the preparation thereof.

⑦ New alkylthiobenzimidazole derivatives are described which belong to the class of formula:



The compounds (I) possess interesting antihistaminic and anti-allergic activities.

wherein R₁ and R₂ each represent a 1-3 carbon atom alkyl radical or they may form with the adjacent nitrogen atom, an optionally substituted heterocyclic ring,
X represents a hydrogen atom or methyl radical
n is 1 or 2

R₃ represents a 4-6 carbon atom alkoxyalkyl radical, a 7 or 8 carbon atom arylalkyl radical or a 5 or 6 carbon atom cycloalkyl radical with the exception that R₃ may not be an arylalkyl radical when R₁ and R₂ each represent a 1-3 carbon atom alkyl radical
R₄ and R₅ may be the same or different and each represent a hydrogen atom or a 1-2 carbon atom alkyl or they, attached at the positions 5 and 6 of the benzimidazole nucleus, may form together the ring

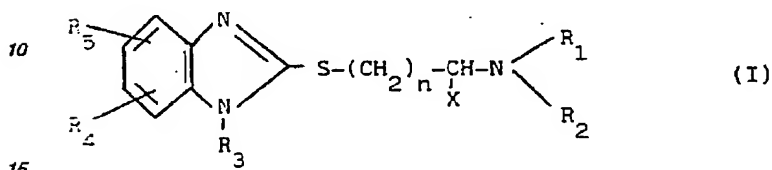
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Description

PHARMACOLOGICALLY ACTIVE ALKYLTHIOBENZIMIDAZOLE DERIVATIVES AND PROCESS FOR THE PREPARATION THEREOF

The present invention refers to a new class of alkylthio benzimidazole derivatives having an interesting antihistaminic and anti-allergic activities and to the process for the preparation thereof.

More particularly, the compounds of the present invention are represented by the class of compounds having the structure formula:



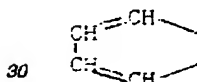
wherein R₁ and R₂ each represent a 1-3 carbon atom alkyl radical or they may form with the adjacent nitrogen atom, an optionally substituted heterocyclic ring, in particular an heterocyclic ring selected from the group consisting of optionally substituted pyrrolidine, piperidine, piperazine and morpholine

X represents a hydrogen atom or methyl radical

n is 1 or 2

R₃ represents a 4 - 6 carbon atom alkoxyalkyl radical, a 7 or 8 carbon atom arylalkyl radical or a 5 or 6 carbon atom cycloalkyl radical with the exception that R₃ may not be an arylalkyl radical when R₁ and R₂ each represent a 1-3 carbon atom alkyl radical

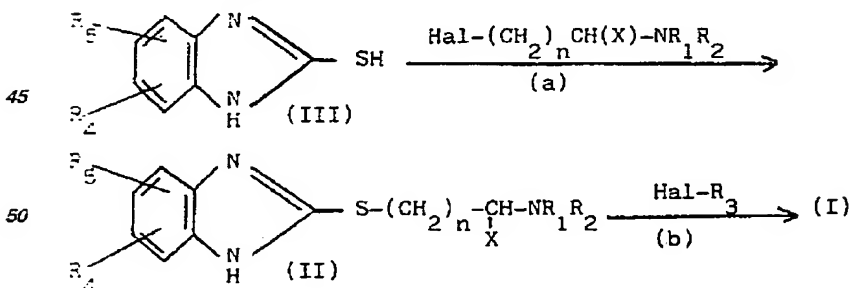
R₄ and R₅ may be the same or different and each represent a hydrogen atom or a 1-2 carbon atom alkyl or they, attached at the positions 5 and 6 of the benzimidazole nucleus, may form together the ring



and by the corresponding non-toxic, pharmaceutically acceptable and suitable acid addition salts.

The compounds of formula (1) are easily prepared starting from 2-mercaptobenzimidazole, which may be substituted at the benzene ring, by first introducing the radical in position 2, by means of reaction in warm conditions, with the suitable aminoalkylhalide Hal-(CH₂)_n-CH(X)-NR₁R₂, wherein X, n, R₁ and R₂ have the above identified meanings and Hal is a halogen atom, preferably chlorine or bromine, then reacting in warm conditions the so obtained 2-thiosubstituted benzimidazole with the halide Hal-R₃, wherein Hal and R₃ have the above meanings.

The process may be schematically described as follows:



wherein R₁, R₂, X, n, R₃, R₄, R₅ and Hal have the above mentioned meanings.

Both the two reactions (a) and (b) are carried out in warm conditions, at a temperature between 50 and 140°C, in an alkaline medium, in suitable organic solvents. The reaction (a) preferably, is carried out in solvents consisting of lower alcohols, preferably ethyl alcohol, while reaction (b) preferably takes place in aprotic solvents, such as dioxane and N,N-dimethylformamide. Both the products (II) and (I), where possible, may be usefully isolated as non-toxic, pharmaceutically acceptable salts with suitable acids.

The anti-histaminic activity of the compounds of the invention has been evaluated studying either the effect on the mortality induced by histamine and the affinity towards H₁ histaminergic receptors. The tests were carried out according to the following methods.

- Effect on the mortality induced by histamine.

The method described by Romer D. et al., (Med.Welt 17, 791,-1966) was followed and the tests were carried out on male albino guinea pigs (Dunkin-Hartley), weighing 350-450g, which were kept in cages with a grid floor, on an empty stomach for 24 hrs and with water ad libitum.

The compounds (I) were dissolved in saline solution and intraperitoneally administered to the animals: 30 minutes later the same animals were intravenously treated with 1,25 mg/kg of histamine dihydrochloride in saline solution. 5

The control animals, to which only the histamine solution had been administered, showed a 100% mortality during the first hour following the treatment.

It was evaluated ID_{50} which corresponds to the amount of the compound able to inhibit 50% of the mortality induced by histamine: the estimation of ID_{50} was carried out by applying the 'probit' method (Finney D.J., "Statistical method in biological assay" pg.512,1957). 10

- Evaluation of the affinity towards H_1 histaminergic receptors.

The affinity of the examined compounds towards H_1 histaminergic receptors was evaluated by displacement curves which were obtained at different concentrations, against [3H]Mepyramine in homogenates of rat brain "in toto", according to the method described by Trau et al. (Proc.Natl.Acad.Sci.,75,6290-1978), with minor modifications. 15

The incubation was performed at 25°C for 15 minutes, in a final volume of 1 ml of 50 mM Na/K phosphate buffer, pH 7.5, in the presence of 2n M [3H]Mepyramine and 1.5 mg of cerebral protein. Aspecific binding was defined as the residual radioactivity bound in the presence of 0.1 mM Clemastine. 20

The test compounds were dissolved in phosphate buffer or dimethylsulfoxide (DMSO), the final concentration of DMSO was 0.5%. Incubation was terminated by filtration through glass fiber GF/B filters pre-soaked with 0.1% polyethylenimine (PEI). The radioactivity trapped by filters was counted by liquid scintillation.

Inhibition constant (KI) was calculated by the method proposed by Cheng and Prusoff (Biochem.Pharmacol.22, 3099-3108, 1973), from IC_{50} values obtained by non-linear fitting analysis of displacement curves. 25

The anti-allergic activity of the compounds of the present invention has been evaluated studying the protection towards the mortality induced by administration of compound 48/80.

- Effect on the mortality induced by compound 48/80.

The tests were carried out according to the method described by C.J.E.Niemergeers et al., Arch.Int.Pharmacodyn.,234,184,-1978. Sprague Dawley Nos male rats (Nossan, Correzzana, Milano) weighing 140-150 g, divided into groups of 10 animals each, on an empty stomach for 24 hrs and with water ad libitum, were kept in cages with a grid floor, then treated intravenously with 2 mg/kg of compound 48/80 (1 ml/rat). The compound (I) or the carrier were dissolved in water (5 ml/kg) and intraperitoneally administered to the animals 30 minutes before the treatment with the compound 48/80. The mortality occurred in the group of treated animals during the first 4 hours following the treatment was noted down. The results were expressed as the number of animals dead with respect to the number of treated animals. 30

The experimental data were submitted to the variance analysis and to the subsequent multiple comparisons according to Dunnett (D.J.Finney, "Statistical Method in Biological Assay", Ed.L.Griffin and Co.,Ltd.,pg.152-157, Edition Ames Iowa,1971). 35

- The Lethal Dose $_{50}$ (LD_{50}) was evaluated on mice using Swiss Nos (Nossan, Correzzana, Milano) mice weighing 18-20 g each. The animals, divided into groups of 10 animals each (5M + 5F), were on an empty stomach for 18 hrs, with water ad libitum, and kept in cages with a grid floor. The compounds (I) were dissolved in water or suspended in 0.5% carboxymethylcellulose and intraperitoneally administered to the animals (10 ml/kg). The mortality occurred in the tested animals within the following 6 hours was noted down. Upon the expiry of the sixth hour, the animals were allowed to eat up to the end of the experimentation which lasted 14 days. During this period all the toxic symptoms and the mortality occurring were noted. 40

The animals which died during the test period and those which were sacrificed at the end of the same, underwent autopsy for a macroscopic examination of their main organs. The experimental data were statistically compared with the X^2 method and LD_{50} was extrapolated by the 'probit' method. 45

The data resulting from the tests carried out on some significant compounds of the class (I), are given in the following Table. The compounds were also tested as to the effect on the sleeping time and it was found that, at their active dose, they practically had no effect on the sleeping time. 50

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Compound	Histamine mortality ID ₅₀ (µg/kg) i.p.	Compound 48/80 mortality ID ₅₀ (µg/kg) i.p.	Sleeping time % increase 25 mg/kg i.p.	Acute toxicity LD ₅₀ (mg/kg) i.p.	H ₁ -receptor binding IC ₅₀ (nM)
Example 3	10.9 (5.5-21.6)	8 (4-15)	6	> 100	2.4
Example 5	50 (19-131)	9 (5-17)	10	~ 100	6.1
Terfenadine	735 (437-1237)	1990 (1080-3720)	8	> 100	283.1

For therapeutic administration, the compounds according to the present invention are used in the form of pharmaceutical preparation which contain said compounds in admixture with pharmaceutically acceptable carriers such as an organic or inorganic solid or liquid excipient suitable for oral or parenteral administration. The compounds of the invention may be contained in these pharmaceutical preparations in the form of free base or in the form of their non-toxic acid addition salts. The inorganic acids which may be employed to prepare these acid addition salts may be, for example, hydrochloric or sulphuric acid. The organic acids which may be employed are, for example, maleic, citric, fumaric and succinic acid. The pharmaceutical preparations may be in solid form as capsules, tablets, dragees or in liquid form such as solutions, suspensions or emulsion. If desired, there may be included in the above preparations auxiliary substances such as stabilizing agent and other commonly used additives, or there may be contained other therapeutically active agents suitable to be administered together with the compounds of the invention. The dosage of the compounds will vary from the administration route and will also depend upon the age and condition of the patient.

The following Examples are given by way of better illustrating the invention, but without limiting it.

Example 1

1-Benzyl-2-[2-(4-methylpiperazin-1-yl)ethylthio]benzimidazole

Grams 2.2 2-mercapto-1H-benzimidazole and 2.4 g NaOH were dissolved in 40 ml 90% ethyl alcohol, then 3.5 g 1-(2-chloroethyl)-4-methylpiperazine hydrochloride were added thereto. The reaction mixture was refluxed for 2 hours, the solid precipitate removed by filtration, the limpid solution evaporated to dryness and the residue dissolved in chloroform and washed with water. The organic phase was made anhydrous, then evaporated to dryness and the residue chromatographed on a column eluting with chloroform, methyl alcohol, cyclohexane, ammonium hydroxide (68:15:15:0.2). The fractions which contained the product, were collected together and evaporated to dryness and the residue which was triturated with isopropyl ether, gave 0.65 g 2-[2-(4-methylpiperazin-1-yl)ethylthio]benzimidazole melting at 93-95°C (with a slight decomposition).

Grams 2 2-[2-(4-methylpiperazin-1-yl)ethylthio] benzimidazole, 0.86 ml benzyl chloride, a 3.5 g aqueous solution containing 1.16 g NaOH and 7.5 ml N,N-dimethylformamide were refluxed for 6 hours, then the solid precipitate was filtered off, and the limpid solution was evaporated almost to dryness. The obtained residue was dissolved in chloroform and water and the extraction was carried out three times; the organic phase was washed, made anhydrous and then evaporated to dryness. The obtained residue was chromatographed on a column eluting with chloroform, methyl alcohol, cyclohexane, ammonium hydroxide (65:15:15:0.1). The fractions, which contained the compound, were evaporated to dryness and the residue which was taken up with boiling petroleum ether (80-100°C) and separated from the insoluble oil, gave, on cooling, 0.3 g 1-benzyl-2-[2-(4-methylpiperazin-1-yl)ethylthio]benzimidazole melting at 122-125°C.

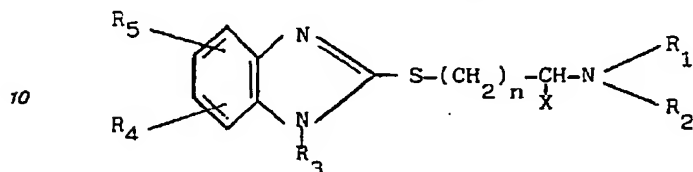
Examples 2-12

Operation was carried out in a manner similar to the above Example and the following compounds were prepared:

- 1-Benzyl-2-[2-(4-chlorobenzhydryl)piperazin-1-yl]ethylthio]benzimidazole, as an oil, starting from 2-[2-(4-chlorobenzhydryl)piperazin-1-yl]ethylthio]benzimidazole (m.p. 98-98°C, with decomposition) and benzyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-dimethylaminoethylthio)benzimidazole, 2 HCl, (m.p. 159-161°C) starting from 2-(dimethylaminoethylthio)benzimidazole and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(3-dimethylaminopropylthio)benzimidazole, as a straw colored oil, starting from 2-(3-dimethylaminopropylthio)benzimidazole (m.p. 77-79°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-dimethylaminoisopropylthio)benzimidazole, starting from 2-(2-dimethylaminoisopropylthio)benzimidazole, (m.p. 166-169°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-piperidin-1-ylethylthio)benzimidazole starting from 2-(2-piperidin-1-ylethylthio)benzimidazole, (m.p. 141-142°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-[2-(N-morpholino)ethylthio]benzimidazole, as an oil (Rf (CHCl₃, CH₃OH 7:3) = 0.82) starting from 2-(2-N-morpholino)ethylthio benzimidazole (m.p. 118-119°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-pyrrolidin-1-ylethylthio)benzimidazole, starting from 2-(2-pyrrolidin-1-ylethylthio)benzimidazole, (m.p. 116-117°C) and 2-ethoxyethyl chloride.
- 1-Cyclopentyl-2-(2-dimethylaminoethylthio)benzimidazole, as an oil, starting from 2-(2-dimethylaminoethylthio)benzimidazole and cyclopentyl bromide.
- 1-(2-Ethoxyethyl)-2-(2-dimethylaminoethylthio)-5,6-dimethyl benzimidazole, as an oil, [Rf (CHCl₃, CH₃OH 7:3) = 0.82], starting from 5,6-dimethyl-2-(2-dimethylaminoethylthio)benzimidazole (m.p. 103-104°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-diethylaminoethylthio)benzimidazole starting from 2-(2-diethylaminoethylthio) benzimidazole (m.p. 104-105°C) and 2-ethoxyethyl chloride.
- 1-(2-Ethoxyethyl)-2-(2-dimethylaminoethylthio)naphtholimidazole starting from 2-(2-dimethylaminoethylthio)naphtholimidazole (m.p. 165-167°C) and 2-ethoxyethyl chloride.

Claims

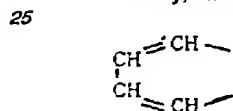
5 1. Alkylthiobenzimidazole derivatives having structure formula:



15 wherein R_1 and R_2 each represent a 1-3 carbon atom alkyl radical or they may form together with the adjacent nitrogen atom, an optionally substituted heterocyclic ring selected from the group consisting of pyrrolidine, piperidine, piperazine and morpholine
 X represents a hydrogen atom or methyl radical
 n is 1 or 2.

20 R_3 represents a 4-6 carbon atom alkoxyalkyl radical, a 7 or 8 carbon atom arylalkyl radical or a 5 or 6 carbon atom cycloalkyl radical with the exception that R_3 may not be an arylalkyl radical when R_1 and R_2 each represent a 1-3 carbon atom alkyl radical

25 R_4 and R_5 may be the same or different and each represent a hydrogen atom or a 1 or 2 carbon atom alkyl or they, attached at the positions 5 and 6 of the benzimidazole nucleus may form together the ring



and the corresponding non-toxic, pharmaceutically acceptable, suitable acid addition salts.

2. Alkylthiobenzimidazole derivatives according to claim 1, wherein R_3 represents 2-ethoxyethyl radical.

3. 1-(2-Ethoxyethyl)-2-(2-dimethylaminoethylthio)benzimidazole.

4. 1-(2-Ethoxyethyl)-2-(2-dimethylaminoisopropylthio)benzimidazole.

35 5. A pharmaceutical composition which comprises a therapeutically active amount of one or more compounds of the claims from 1 to 4 in an admixture with suitable pharmaceutically acceptable diluents.

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European Patent
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EUROPEAN SEARCH REPORT

Application Number

EP 89 83 0122

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl. 4)
A	PATENT ABSTRACTS OF JAPAN, vol. 9, no. 309 (C-318)[2032], 5th December 1985; & JP-A-60 149 569 (KIYOURIN SEIYAKU K.K.) 07-08-1985		C 07 D 235/28 A 61 K 31/415 C 07 D 235/02
A	FR-A-2 232 315 (E.R. SQUIBB & SONS)		
A	EP-A-0 174 717 (FISONS PLC)		
			TECHNICAL FIELDS SEARCHED (Int. Cl. 4)
			C 07 D 235/00 A 61 K 31/00
The present search report has been drawn up for all claims			
Place of search THE HAGUE		Date of completion of the search 30-06-1989	Examiner DE BUYSER I.A.F.
CATEGORY OF CITED DOCUMENTS			
X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document	

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